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FILE 'HOME' ENTERED AT 18:44:28 ON 17 OCT 2006

=> file .nash
=> s thermostable and protease and (peptide bond or peptide ligation)
L1             4 FILE MEDLINE
L2            15 FILE CAPLUS
L3             6 FILE SCISEARCH
L4             3 FILE LIFESCI
L5             6 FILE BIOSIS
L6             4 FILE EMBASE

TOTAL FOR ALL FILES
L7            38 THERMOSTABLE AND PROTEASE AND (PEPTIDE BOND OR PEPTIDE LIGATION)

=> s l7 not 1997-2006/py
L8             2 FILE MEDLINE
L9             9 FILE CAPLUS
L10            2 FILE SCISEARCH
L11            2 FILE LIFESCI
L12            4 FILE BIOSIS
L13            2 FILE EMBASE

TOTAL FOR ALL FILES
L14           21 L7 NOT 1997-2006/PY

=> dup rem
ENTER L# LIST OR (END):l14
PROCESSING COMPLETED FOR L14
L15            9 DUP REM L14 (12 DUPLICATES REMOVED)

=> d ibib abs 1-9

L15  ANSWER 1 OF 9  CAPLUS  COPYRIGHT 2006 ACS on STN  DUPLICATE 1
ACCESSION NUMBER:         1996:502382  CAPLUS  Full-text
DOCUMENT NUMBER:          125:161800
TITLE:                    An extremely thermostable serine
                          protease from a hyperthermophilic archaeum,
                          Desulfurococcus strain SY, isolated from a deep-sea
                          hydrothermal vent
AUTHOR(S):                Hanzawa, Satoshi; Hoaki, Toshihiro; Jannasch, Holger
                          W.; Maruyama, Tadashi
CORPORATE SOURCE:         Shimizu Laboratories, Marine Biotechnology Institute,
                          Shimizu, 424, Japan
SOURCE:                   Journal of Marine Biotechnology (1996), 4(2), 121-126
                          CODEN: JMBOEW; ISSN: 0941-2905
PUBLISHER:                Springer
DOCUMENT TYPE:            Journal
LANGUAGE:                 English
AB      An extremely thermostable serine protease was purified from a deep-sea hyperthermophilic, sulfur-
        dependent, heterotrophic archaeum, Desulfurococcus strain SY, which requires 11 amino acids for
        growth.  Protease activity was detected in cell suspensions only, and not in the medium after the
        removal of cells by centrifugation. Mol. weight of the enzyme was 240 kDa as determined by gel-
        filtration high-pressure liquid chromatog. (HPLC), while it was 70 kDa by sodium dodecyl sulfate-
        polyacrylamide gel electrophoresis (SDS-PAGE).  Optimum temperature of the activity on azocasein
        was 105°C at pH 7.5.  The protease was most stable at pH 5.9.  The activity increased 2-30% after
        0.5-1 h of incubation at 90-98°C and then decreased exponentially with a half-life (at pH 5.9) of
        9.1, 4.3, and 2.5 h at 90, 95, and 98°C, resp.  Optimum pH values for the activity on azocasein,
        succinyl-Ala-Pro-Phe-p-nitroanilide, and succinyl-Ala-Ala-Val- Ala-p-nitroanilide were 8, 7, and
        6, resp.  Optimum NaCl concentration for the activity on azocasein was 0.5 M but was 2-4 M on
        succinyl-Ala-Ala-Val-Ala- p-nitroanilide.  The enzyme cleaved peptide bonds of proangiotensin at
        Tyr5-Ile6 and Phe8-His9 and of oxidized insulin B chain at Leu15-Tyr16, Leu17-Val18, and Phe25-
        Tyr26.


L15  ANSWER 2 OF 9      MEDLINE on STN                    DUPLICATE 2
ACCESSION NUMBER:    93107026      MEDLINE  Full-text
DOCUMENT NUMBER:     PubMed ID: 8416942
TITLE:               Purification and characterization of kumamolysin, a novel
                     thermostable pepstatin-insensitive carboxyl
                     proteinase from Bacillus novosp. MN-32.
AUTHOR:              Murao S; Ohkuni K; Nagao M; Hirayama K; Fukuhara K; Oda K;
                     Oyama H; Shin T
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CORPORATE SOURCE: Department of Applied Microbial Technology, Kumamoto Institute of Technology, Japan.
SOURCE: The Journal of biological chemistry, (1993 Jan 5) Vol. 268, No. 1, pp. 349-55.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199301
ENTRY DATE: Entered STN: 12 Feb 1993
Last Updated on STN: 3 Mar 2000
Entered Medline: 28 Jan 1993

AB We have found a novel type of thermostable, pepstatin- insensitive carboxyl proteinase in the culture filtrate of Bacillus novosp. MN-32. The carboxyl proteinase, which was named kumamolysin, was purified about 8,300-fold by column chromatography including DEAE-Sepharose CL-6B, Sephadex G-100, and TSKgel DEAE-5PW. The purified kumamolysin gave a single band corresponding to 41 kDa by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. The molecular mass of kumamolysin was estimated to be 40 kDa by gel filtration. The isoelectric point of kumamolysin was estimated to be pH 3.5 by isoelectric focusing. Kumamolysin has maximum proteolytic activity at 70 degrees C and at pH 3.0. Kumamolysin specifically hydrolyzed the Leu15-Tyr16 peptide bond in oxidized insulin B-chain (Km = 9.0 x 10(-5) M, Kcat = 71 s-1; at pH 3.0, 30 degrees C), and additional cleavage at Phe25-Tyr26 was detected at a considerably lower rate. Kumamolysin is insensitive to the known carboxyl proteinase inhibitors pepstatin, diazoacetyl-DL-norleucine methyl ester, and 1,2-epoxy-3-(p-nitrophenoxy)propane. Kumamolysin has no similarity to the thermostable acid protease thermopsin from Sulfolobus acidocaldarius (Lin, X.-L., and Tang, J. (1990) J. Biol. Chemical 265, 1490-1495). Thus, the substrate specificity, the inhibitor sensitivity, the molecular mass, and the thermostability all suggest that kumamolysin is a novel type of carboxyl proteinase.

L15 ANSWER 3 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1993:192266 CAPLUS Full-text
DOCUMENT NUMBER: 118:192266
TITLE: Some characteristics of a serine proteinase isolated from an extreme thermophile for use in kinetically controlled peptide bond synthesis
AUTHOR(S): Peek, Keith; Wilson, Shelley Ann; Prescott, Mark; Daniel, Roy M.
CORPORATE SOURCE: Pac. Enzymes Ltd., Hamilton, N. Z.
SOURCE: Annals of the New York Academy of Sciences (1992), 672(Enzyme Engineering XI), 471-7
CODEN: ANYAA9; ISSN: 0077-8923
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Optimization of peptide coupling of Bz-Ala-OMe with H-Tyr-NH2 catalyzed by a serine proteinase isolated from Thermus sp. strain Rt41A is described.

L15 ANSWER 4 OF 9 MEDLINE on STN DUPLICATE 3
ACCESSION NUMBER: 92111476 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 1765065
TITLE: The conformational stability of the redox states of lipoamide dehydrogenase from Azotobacter vinelandii.
AUTHOR: van Berkel W J; Regelink A G; Beintema J J; de Kok A
CORPORATE SOURCE: Department of Biochemistry, Agricultural University, Wageningen, The Netherlands.
SOURCE: European journal of biochemistry / FEBS, (1991 Dec 18) Vol. 202, No. 3, pp. 1049-55.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199202
ENTRY DATE: Entered STN: 8 Mar 1992
Last Updated on STN: 3 Feb 1997
Entered Medline: 14 Feb 1992

AB The conformational stability of holo-lipoamide and apo-lipoamide dehydrogenase from Azotobacter vinelandii was studied by thermoinactivation, unfolding and limited proteolysis. The oxidized holoenzyme is thermostable, showing a melting temperature, tm = 80 degrees C. The thermal stability of the holoenzyme drastically decreases upon reduction. Unlike the oxidized and lipoamide two-electron reduced enzyme species, the NADH four-electron reduced enzyme is highly sensitive to unfolding by urea. Loss of energy transfer from Trp199 to flavin reflects the unfolding of the oxidized holoenzyme by guanidine hydrochloride. Unfolding of the monomeric

apoenzyme is a rapid fully reversible process, following a simple two-state mechanism. The oxidized and two-electron reduced holoenzyme are resistant to limited proteolysis by trypsin and endoproteinase Glu-C. Upon cleavage of the apoenzyme or four-electron reduced holoenzyme by both proteases, large peptide fragments (molecular mass greater than 40 kDa) are transiently produced. Sequence studies show that limited trypsinolysis of the NADH-reduced enzyme starts mainly at the C-terminus of Arg391. In the apoenzyme, limited proteolysis by endoproteinase Glu-C starts from the C-terminus at the carboxyl ends of Glu459 and/or Glu435. From crystallographic data it is deduced that the susceptible amino acid peptide bonds are situated near the subunit interface. Thus, these bonds are inaccessible to the proteases in the dimeric enzyme and become accessible after monomerization. It is concluded that reduction of lipoamide dehydrogenase to the four-electron reduced state(s) is accompanied by conformational changes promoting subunit dissociation.

L15 ANSWER 5 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN DUPLICATE 4
ACCESSION NUMBER: 1988:488549 CAPLUS Full-text
DOCUMENT NUMBER: 109:88549
TITLE: Thermostable alkaline protease produced by Bacillus thermoruber - a new species of Bacillus
AUTHOR(S): Manachini, Pier L.; Fortina, Maria G.; Parini, Carlo
CORPORATE SOURCE: Dip. Sci. Tecnol. Alimentari Microbiol., Univ. Studi Milano, Milan, I-20133, Italy
SOURCE: Applied Microbiology and Biotechnology (1988), 28(4-5), 409-13
CODEN: AMBIDG; ISSN: 0175-7598
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The proteolytic activity produced by a new species of Bacillus isolated in the authors' laboratory was investigated. Thermostable alkaline protease was purified to homogeneity from cell-free culture liqs. of B. thermoruber. The purification procedure included ion-exchange chromatog. on DEAE-Sephadex A-50 and α-casein agarose affinity chromatog. The protease consisted of 1 polypeptide chain with a mol. weight of 39,000. The pI was 5.3; proteolytic activity (on casein) was optimum at pH 9 and 45°, resp. Enzyme activity was inhibited by phenylmethanesulfonyl fluoride and EDTA. The stability of the enzyme was considerably increased by addition of Ca2+, and the protease exhibited a relatively high thermal stability. The alkaline (serine) protease showed a preference for leucine in the carboxylic side of the peptide bond of the substrate. The Km for benzyloxycarbonyl-Ala-Ala-Leu-p-nitroanilide was 2.5 mM.

L15 ANSWER 6 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1975:151107 CAPLUS Full-text
DOCUMENT NUMBER: 82:151107
TITLE: Proteolytic system of Penicillium roqueforti. III. Purification, properties, and specificity of a protease inhibited by EDTA
AUTHOR(S): Gripon, Jean C.; Hermier, Jean
CORPORATE SOURCE: Cent. Natl. Rech. Zool., Inst. Natl. Rech. Agron., Jouy-en-Josas, Fr.
SOURCE: Biochimie (1974), 56(10), 1323-32
CODEN: BICMBE; ISSN: 0300-9084
DOCUMENT TYPE: Journal
LANGUAGE: French
AB The extracellular protease (I) of P. roqueforti was isolated from the medium by (NH4)2SO4 precipitation, gel filtration, and CM-cellulose chromatog. I differed from the acid protease of the mold. Purified I was homogenous on polyacrylamide gel electrophoresis at pH 9.2, 9.0, and 4.3. Mol. weight was estimated as 20,000 daltons by gel filtration. PH optima for casein and hemoglobin digestion were 5.5 and 4.2, resp. I was thermostable after 20 min at 100°, residual activity was still 25% (pH 6). I was very unstable at 65°. Co2+ promoted casein hydrolysis. Chelating agents inhibited I completely. Extracellular I had an entirely different specificity from that found for neutral I. It did not split disubstituted peptides which are the classical substrates for neutral I. It rapidly hydrolyzed 2 peptide bonds on the oxidized insulin B-chain, but did not attack other peptide bonds which are usually hydrolyzed by metalloproteases (e.g., Bacillus megaterium). Beef trypsinogen, collagen, keratin, fibrin, gelatin, or elastin was not hydrolyzed. The basic proteins, histone and protamine, were hydrolyzed 2- to 3-fold more rapidly than casein.

L15 ANSWER 7 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1967:72703 CAPLUS Full-text
DOCUMENT NUMBER: 66:72703
TITLE: Comparison of substrate specificities of a thermostable bacterial proteinase and a bacterial elastase
AUTHOR(S): Morihara, Kazuyuki; Ebata, Mitsuo
CORPORATE SOURCE: Shionogi Co., Ltd., Osaka, Japan

SOURCE: Journal of Biochemistry (Tokyo, Japan) (1967), 61(1), 149-51
CODEN: JOBIAO; ISSN: 0021-924X
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The action of a heat-stable proteinase from Bacillus thermoproteolyticus and an elastase from Pseudomonas aeruginosa was determined on 11 synthetic peptides. Both enzymes had the same substrate specificity, hydrolyzing the peptide bonds at the amino end of leucine and phenylalanine residues of L-configuration. The Km values and maximum velocities of the enzymes were also similar.

L15 ANSWER 8 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1966:449270 CAPLUS Full-text
DOCUMENT NUMBER: 65:49270
ORIGINAL REFERENCE NO.: 65:9256f-g
TITLE: Specific nature of hydrolysis of insulin and tobacco mosaic virus protein by thermolysin
AUTHOR(S): Matsubara, H.; Sasaki, R.; Singer, A.; Jukes, T. H.
CORPORATE SOURCE: Univ. of California, Berkeley
SOURCE: Archives of Biochemistry and Biophysics (1966), 115(2), 324-31
CODEN: ABBIA4; ISSN: 0003-9861
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Oxidized bovine insulin and tobacco mosaic virus protein were used as substrates in an investigation of the specificity of thermolysin, a thermostable protease. Under the conditions used, thermolysin hydrolyzed the peptide bonds involving the amino groups of the leucine and phenylalanine residues of insulin, and preferentially those of the leucine, isoleucine, valine, and phenylalanine residues of tobacco mosaic virus protein. It was concluded that thermolysin hydrolyzes preferentially the peptide bonds involving the amino groups of hydrophobic amino acid residues with bulky side chains. 17 references.

L15 ANSWER 9 OF 9 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1966:20945 CAPLUS Full-text
DOCUMENT NUMBER: 64:20945
ORIGINAL REFERENCE NO.: 64:3908f-g
TITLE: Observations on the specificity of a thermostable bacterial protease: thermolysin
AUTHOR(S): Matsubara, Hiroshi; Singer, Alan; Sasaki, Richard; Jukes, Thomas H.
CORPORATE SOURCE: Univ. of California, Berkeley
SOURCE: Biochemical and Biophysical Research Communications (1965), 21(3), 242-7
CODEN: BBRCA9; ISSN: 0006-291X
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The substrate specificity of thermolysin was studied using beef heart cytochrome c as a substrate. The peptides produced were separated by 2-dimensional paper chromatography and electrophoresis, and analyzed. Thermolysin hydrolyzed 10 of the 12 peptide bonds involving the NH2 groups of isoleucine and leucine residues in cytochrome c under the exptl. conditions. Three other bonds, involving the amino sites of phenylalanine and alanine residues, were also hydrolyzed. The property of thermolysin in preferentially attacking leucine and isoleucine residues in peptide linkage is of potential usefulness in studies of the primary structure of proteins.

=> s thermostable and (protease or proteinase) and (bond formation or peptide ligation)
L16 1 FILE MEDLINE
L17 1 FILE CAPLUS
L18 1 FILE SCISEARCH
L19 0 FILE LIFESCI
L20 1 FILE BIOSIS
L21 1 FILE EMBASE

TOTAL FOR ALL FILES
L22 5 THERMOSTABLE AND (PROTEASE OR PROTEINASE) AND (BOND FORMATION OR PEPTIDE LIGATION)

=> dup rem l22
PROCESSING COMPLETED FOR L22
L23 1 DUP REM L22 (4 DUPLICATES REMOVED)

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L23 ANSWER 1 OF 1 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2004295431 MEDLINE Full-text
DOCUMENT NUMBER: PubMed ID: 15196009
TITLE: Generation of a thermostable and denaturant-resistant peptide ligase.
AUTHOR: Joe Koman; Borgford Thor J; Bennet Andrew J
CORPORATE SOURCE: Department of Molecular Biology and Biochemistry, Simon Fraser University, 8888 University Drive, Burnaby, British Columbia, Canada V5A 1S6.
SOURCE: Biochemistry, (2004 Jun 22) Vol. 43, No. 24, pp. 7672-7. Journal code: 0370623. ISSN: 0006-2960.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200408
ENTRY DATE: Entered STN: 16 Jun 2004
Last Updated on STN: 10 Aug 2004
Entered Medline: 9 Aug 2004

AB The construction and characterization of a novel, thermostable, peptide ligase are described. Three amino acid substitutions were introduced into the secreted bacterial protease Streptomyces griseus protease B (SGPB). Mutations were chosen on the basis of two separate observations: (i) that a single substitution of the nucleophilic serine (S195A) created an enzyme with significant peptide-ligation activity, albeit greatly reduced stability [(2000) Chemical Biol. 7, 163], and (ii) that a pair of substitutions in the substrate-binding pocket (T213L and F228H) greatly increased the thermostability of the wild-type enzyme [(1996) J. Mol. Biol. 257, 233]. The triple mutant, named streptoligase, was found to catalyze peptide ligation (aminolysis of both a thiobenzyl ester and a p-nitroanilide-activated peptide) efficiently in nondenaturing and denaturing conditions including SDS (0.5% w/v) and guanidine hydrochloride (4.0 M). Moreover, streptoligase exhibited a half-live for unfolding of 16.3 min at 55 degrees C in the absence of stabilizing substrates. The fraction of the streptoligase-catalyzed reaction that gave coupled product with the acceptor peptide FAASR-NH(2) was greater for the p-nitroanilide donor (Sc-AAPF-pNA) than for the benzyl thioester substrate (Sc-AAPF-SBn). These observations are consistent with ligation proceeding through an acyl-enzyme intermediate involving histidine-57. In the case of the thioester donor the triple mutant promotes the direct attack of water on the thioester carbonyl carbon, in addition to hydrolysis occurring at the stage of the acyl-enzyme intermediate. The strategy of multiple point mutations outlined in this study may provide a general means of converting enzymes with chymotrypsin-like protein folds into peptide ligases.

=> log y